Automated headspace solid-phase dynamic extraction to analyse the volatile fraction of food matrices

Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Chromatography A, 1024 (2004) 217–226
JOURNAL OF CHROMATOGRAPHY A
www.elsevier.com/locate/chroma

Automated headspace solid-phase dynamic extraction to analyse the volatile fraction of food matrices

Carlo Bicchi*, Chiara Cordero, Erica Liberto, Patrizia Rubiolo, Barbara Sgorbini

Dipartimento di Scienza e Tecnologia del Farmaco, Università degli studi Torino, Via Pietro Giuria 9, I-10125 Torino, Italy

Received 11 July 2003; received in revised form 2 October 2003; accepted 8 October 2003

Abstract

High concentration capacity headspace techniques (headspace solid-phase microextraction (HS-SPME) and headspace sorptive extraction (HSSE)) are a bridge between static and dynamic headspace, since they give high concentration factors as does dynamic headspace (D-HS), and are as easy to apply and as reproducible as static headspace (S-HS). In 2000, Chromtech (Idstein, Germany) introduced an inside-needle technique for vapour and liquid sampling, solid-phase dynamic extraction (SPDE), also known as "the magic needle". In SPDE, analytes are concentrated on a 50 μm film of polydimethylsiloxane (PDMS) and activated carbon (10%) coated onto the inside wall of the stainless steel needle (5 cm) of a 2.5 ml gas tight syringe. When SPDE is used for headspace sampling (HS-SPDE), a fixed volume of the headspace of the sample under investigation is sucked up an appropriate number of times with the gas tight syringe and an analyte amount suitable for a reliable GC or GC–MS analysis accumulates in the polymer coating the needle wall. This article describes the preliminary results of both a study on the optimisation of sampling parameters conditioning HS-SPDE recovery, through the analysis of a standard mixture of highly volatile compounds (β-pinene, isoamyl acetate and linalool) and of the HS-SPDE–GC–MS analyses of aromatic plants and food matrices. This study shows that HS-SPDE is a successful technique for HS-sampling with high concentration capability, good repeatability and intermediate precision, also when it is compared to HS-SPME.

Keywords: Extraction methods; Solid-phase dynamic extraction; Headspace analysis; Food analysis; Automation; Volatile organic compounds; Aroma compounds

1. Introduction

Headspace sampling is a solvent-free technique used in combination with GC and GC–MS to characterize the volatile fraction of several matrices including food matrices and aromatic and medicinal plants. Over the last 10 years, there has been a remarkable renewal of interest in headspace sampling in particular after the introduction of high concentration capacity techniques (HCC) in which the recovery of volatile is mainly based on the sorption approach. The first HCC headspace sampling technique was solid-phase microextraction (HS-SPME), developed and applied to HS sampling by Zhang and Pawliszyn in 1993 [1]. Table 1 reports a list of abbreviations and acronyms. HS-SPME has been shown to be a successful bridge between static (S-HS) and dynamic (D-HS) headspace being as simple, as reproducible and as easy to automate as S-HS, and as sensitive (because of the analyte enrichment achieved with the fibre) and as selective (because of different absorption characteristics of the fibre coating constituents) as D-HS. One of the limits of SPME is its reduced concentration capability, which is mainly due to the small volume of polymer coating the fibre (V_{PDMS} in a PDMS 100 μm fibre: about 0.6 μl). This limit was recently overcome by a new technique that is an extension of stir bar sorptive extraction (SBSE) [2]. Headspace sorptive extraction (HSSE) was introduced by Tienpont et al. [3] and Bicchi et al. [4] and is characterised by a high concentration capacity that is mainly due to the high volume of PDMS coating the stir bars (from 25 to 200 μl). Both techniques have shown themselves to be successful in several applications in particular food matrices and aromatic and medicinal plants [4–6].

Several attempts to overcome some other minor disadvantages of SPME (fragility of the fused-silica, unprotected sta-

* Corresponding author. Tel.: +39-011-670-7662; fax: +39-011-670-7687.
E-mail address: carlo.bicchi@unito.it (C. Bicchi).

0021-9673/$ – see front matter © 2003 Elsevier B.V. All rights reserved.
doi:10.1016/j.chroma.2003.10.009

Table 1
List of abbreviations and acronyms and meanings

Acronym or abbreviation	
CF	Concentration factor
D-HS	Dynamic headspace
HCC	High concentration capacity techniques
HSSE	High capacity headspace sorptive extraction
HS-SPDE	Headspace solid-phase dynamic extraction
HS-SPME	Headspace solid-phase microextraction
HTS-FSOT capillary column	High temperature silylation-fused silica open tubular capillary column
PDMS	Polydimethylsiloxane
PEG-20M	Polyethylene glycol 20M
SBSE	Stir bar sorptive extraction
S-HS	Static headspace
SPDE	Solid-phase dynamic extraction
SPME	Solid-phase microextraction
β	Phase ratio

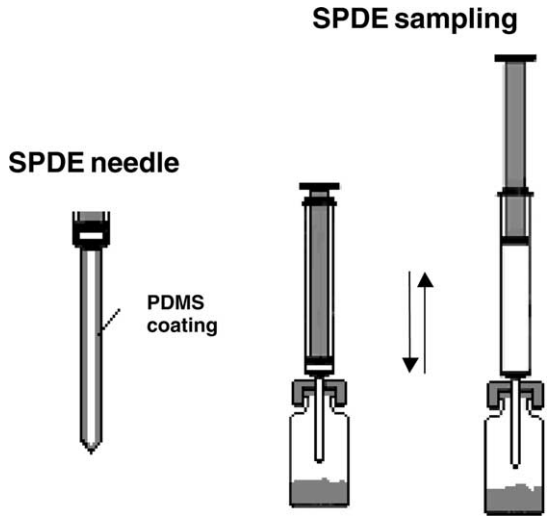

Fig. 1. HS-SPDE sampling device and HS-SPDE PDMS needle.

tionary phase coating, limited flexibility of surface area and film thickness) while keeping constant its great concentration capacity, high extraction speed and stability have been made by coating the inside of a needle or a capillary instead of the outside of a fibre [7,8]. In particular, in 1997 Eisert and Pawliszyn successfully introduced in-tube SPME–LC where sampling was through an open tubular fused-silica capillary column [9].

In 2000, Chromtech (Idstein, Germany) introduced solid-phase dynamic extraction (SPDE), an inside-needle technique for vapour and liquid sampling. SPDE is also known as "the magic needle". In SPDE, the analytes are concentrated on a 50 μm film of polydimethylsiloxane (PDMS) and activated carbon (10%) coated onto the inside wall of the stainless steel needle (5.5 cm long) of a gas tight syringe (2.5 ml). When used for HS-SPDE, an analyte amount sufficient for a reliable GC or GC–MS analysis is accumulated in the polymer coating of the inside needle wall of the gas tight syringe by pulling in and pushing out a fixed volume of the headspace of the sample under investigation for an appropriate number of times. The SPDE sampling allows us to operate under dynamic conditions while keeping constant the headspace volume. The trapped analytes are then recovered by heat desorption directly into a GC injector body. The diagram in Fig. 1 shows SPDE sampling and needle. The volume of PDMS coated on the SPDE needle wall is about 4.5 μl in comparison to about 0.6 μl coating a PDMS 100 μm SPME fibre. To date, SPDE has been successfully applied to the analysis of pesticides in water, by Lipinski [10], and amphetamines in hair samples of drug abusers by Musshoff et al. [11]. This article reports the preliminary results of both a study to optimise HS-SPDE sampling parameters to maximise recovery by analysing a standard mixture of highly volatile compounds and of HS-SPDE–GC–MS analyses applied to food matrices and aromatic plants.

2. Experimental

2.1. Matrices and chemicals

Samples of commercially available dried rosemary leaves (*Rosmarinus officinalis* L.), green and roasted coffee, white and red wines and fresh banana fruits were used. Standard solutions of β-pinene (6.6 μM), isoamyl acetate (1.92 μM) and linalool (4.86 μM) (Sigma–Aldrich, Milan, Italy) in water were also prepared. HPLC grade water was further purified by stirring under vacuum (1×10^{-3} bar) at 60 °C for 2 h before use to avoid interference of the volatile impurities with the analytes under investigation.

2.2. S-HS, HS-SPME and HS-SPDE–GC–MS analysis

2.2.1. HS-SPDE system and sampling

The SPDE equipment (syringes with attached SPDE needles and SPDE gas station) was by Chromtech (Idstein, Germany) and it was installed in a CTC-Combi-PAL-Autosampler (Bender and Hobein, Zurich, Switzerland) in its turn assembled on a GC–MS system consisting of an Agilent model 6890 Series Plus/5973 N. The CTC-Combi-PAL-Autosampler (Bender and Hobein) included an incubator oven with one heated vial position and shaker (Agitator) (Chromtech). All SPDE sampling steps were automatically controlled by the CTC-Combi-PAL software. The SPDE needle (50 mm × 0.8 mm, i.d. 0.53 mm, conical needle tip with side port) coated inside with PDMS containing 10% of activated carbon is assembled onto a 2.5 ml gas-tight syringes with a side port for gas flushing (Hamilton, Darmstadt, Germany). Gas station and syringe are connected to helium for flushing and controlled by the autosampler. The gas station is to supply a fixed volume of helium for desorption. The side port of the syringe cannot be used for desorption, because it has no pressure regulator. The syringe adapter heater was set at 55 °C.

2.2.1.1. Standard mixtures. The influence of some operative parameters on HS-SPDE recovery was evaluated through a series of analyses of a standard mixture of β-pinene (6.6 μM), isoamyl acetate (1.92 μM) and linalool (4.86 μM) in HPLC grade water. In particular, the sampling temperature, number of aspiration cycles, plunger speed and aspired volume for each cycle, helium desorption volume, desorption plunger speed and total volume of sampled headspace were tested (see Section 3). Sampling conditions and procedure are reported in Section 2.2.1.2. Each experiment was repeated three times. Blank runs were carried out, but no carry over was observed.

2.2.1.2. Real world samples. HS-SPDE sampling conditions were chosen on the basis of the results of the experiments carried out on the standard mixture (Section 2.2.1.1). A volume of 2 ml of the matrices investigated were introduced in 21.2 ml vials and hermetically sealed in order to obtain a phase ratio of 9.6. The resulting samples were all equilibrated at 50 °C for 15 min., with the exception of fresh banana, which was sampled at 35 °C. Headspace was sampled by HS-SPDE under the following conditions: agitator (sampling) temperature: 50 °C; headspace syringe temperature: 55 °C; number of filling cycles per extraction: 50; plunger speed for extraction: 50 μl/s (each aspiration taking 40.5 s); helium volume for desorption: 1 ml; plunger speed for desorption: 15 μl/sec; pre-desorption time in the GC injection port: 30 s; desorption temperature: 230 °C. The trapped analytes were recovered by thermal desorption from the SPDE needle directly into the GC injector body and analysed by GC–MS under the conditions reported below (Section 2.2.2). Each experiment was repeated three times; six experiments per day for 3 days were run with rosemary to evaluate repeatability and intermediate precision. Blank runs were carried out, but no carry over was observed.

2.2.2. S-HS and HS-SPME and sampling

S-HS and HS-SPME samplings were carried out automatically with the same instrumentation described above (see Section 2.2.1). For HS-SPME a special holder was installed in the assembly. Polydimethylsiloxane 100 μm fibre (PDMS 100) was used for HS-SPME sampling (Supelco, Bellefonte, PA, USA).

Sampling conditions for both series of experiments were exactly the same as those reported for HS-SPDE. An equilibration time of 60 min was used for S-HS, 1 ml of the vapour phase was directly injected into the GC system; for HS-SPME, the PDMS 100 fibre was exposed to the headspace of the matrices for 60 min.

Before sampling, the PDMS 100 fibre was reconditioned for 30 min in the GC injection port at 230 °C. Each experiment was repeated three times. Six experiments per day for 3 days were run with rosemary to determine HS-SPME repeatability and intermediate precision.

2.2.3. GC–MS analysis

GC–MS analyses were carried out on a HTS-FSOT capillary column (PEG 20M, 25 m × 0.25 mm i.d., film thickness 0.25 μm, MEGA (Legnano, Italy)). Oven temperature programmes were chosen in order to obtain the most effective separation of the headspace components of each matrix investigated and are reported in the captions to figures. Injector temperature: 230 °C, mode splitless; transfer line: 250 °C. Carrier gas: helium, flow-rate: 1.0 ml/min. MS was in the electron impact ionization mode at 70 eV. The HS components were identified by comparison of their mass spectra with those of authentic samples or with data in the literature.

3. Results and discussion

The main difference between headspace sampling by HS-SPDE and HS-SPME and HSSE is that the former is a non-equilibrium sampling method close to the dynamic headspace approach while the latter two are equilibrium sampling methods close to the static headspace approach. HS-SPDE must therefore be carried out under rigorous standard conditions to obtain reliable results in particular for quantitative analysis, since the composition of the fraction sampled by HS-SPDE is strongly influenced by sampling conditions and matrix effect.

A series of experiments was carried out to find the optimal SPDE sampling conditions to maximise recovery. Sampling temperature, number of aspiration cycles, plunger speed and volume aspired for each cycle, total volume of sampled headspace, helium desorption volume and plunger speed for desorption were investigated by analysing a standard mixture consisting of β-pinene (6.6 μM), isoamyl acetate (1.92 μM) and linalool (4.86 μM) in HPLC grade water. These compounds were chosen because of their different physico-chemical properties (see Table 2). Moreover, a phase ratio of 9.6 was arbitrarily chosen so that the volume aspired for each cycle (1 ml) was about 5%

Table 2
Physico-chemical properties of standard mixture components

Compounds	M_r	$\log K_{o/w}$	Vapour pressure (Pa)	Henry law constant (atm m^3/mol)	Analyte concentration (μM)
β-Pinene	136.24	4.35	**390.10**	9.20×10^{-2}	6.60
Isoamyl acetate	130.19	2.26	**746.76**	9.08×10^{-4}	1.92
Linalool	154.25	3.38	**21.28**	2.47×10^{-5}	4.86

1atm = 101 325 Pa.

Table 3
List and values of the HS-SPDE sampling parameters optimised using the β-pinene, isoamyl acetate and linalool standard mixture

Temperature (°C)	Number of aspiration cycles	Aspiration plunger speed (μl/s)	Volume aspiration cycle (ml)	Helium desorption volume (ml)	Desorption plunger speed (μl/s)
30	10	25	0.5	0.5	10
50	**50**	**50**	**1.0**	**1.0**	**15**
70	100	75	2.0	2.0	25
	150	100			

The parameters producing the highest recoveries are in bold.

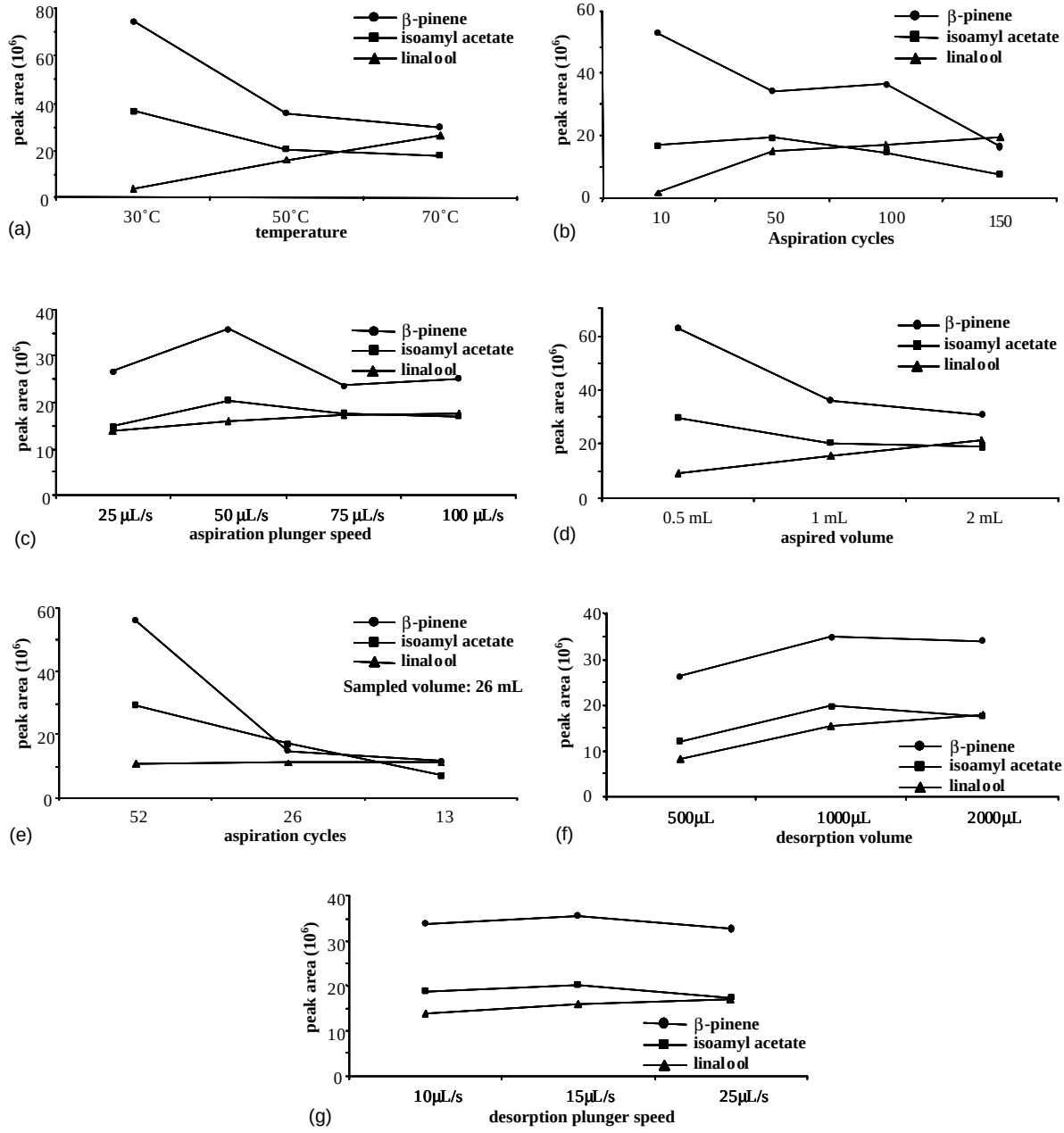

Fig. 2. Optimization of HS-SPDE sampling parameters with a standard mixture of β-pinene (6.6 μM), isoamyl acetate (1.92 μM) and linalool (4.86 μM) in HPLC grade water. (a) Sampling temperature, (b) number of aspiration cycles, (c) plunger speed, (d) volume aspired for each cycle, (e) aspiration cycles with a fixed total volume of sampled headspace, (f) helium desorption volume and (g) plunger speed for desorption. GC oven temperature: from 20 °C (1 min) to 60 °C at 40 °C/min then to 100 °C at 10 °C/min then to 220 °C (5 min) at 20 °C/min (for further detail see text).

of the volume of the gas phase and should not interfere (or only moderately interferes) with the headspace equilibrium.

3.1. Sampling temperatures

The influence of different temperatures (30, 50, and 70 °C) on HS-SPDE recovery of the standard sample was then evaluated under standardised conditions (Table 3). Fig. 2a reports the peak areas of each analyte in function of temperature. β-Pinene and isoamyl acetate behave similarly since their peak areas halved from 30 to 50 °C while that of linalool increased constantly by increasing the temperature. A temperature of 50 °C was therefore chosen for all the remaining experiments as a compromise in consideration of the different polarity and volatility of the components of both standard mixture and real-world matrices investigated.

3.2. Number of aspiration cycles

A set of standard samples was submitted to HS-SPDE under standardised conditions (Table 3) while applying different numbers of aspiration cycles (10, 50, 100, and 150 for a total sampled volume of 10, 50, 100, and 150 ml, respectively). Fig. 2b reports the peak areas of each analyte in function of the number of aspiration cycles. These results clearly show that a good compromise in terms of recovery for all analytes and sampling times can be obtained with 50 cycles because with the increasing of the number of cycles the recovery of linalool increases while that of β-pinene and isoamyl acetate slightly decreases. Of course, the number of aspiration cycles also depends on the headspace volume to be sampled.

3.3. Aspiration plunger speed

This parameter was evaluated by analysing a set of standard samples under standardised conditions (Table 3), but with different plunger speed: 25, 50, 75 and 100 μl/s. Fig. 2c reports the peak areas of each analyte in function of the plunger speed and shows that a plunger speed of 50 μl/sec affords a good recovery of all analytes, in particular for β-pinene and isoamyl acetate.

3.4. Volume of a single aspiration cycle

HS-SPDE was applied to a set of standard samples under standardised conditions (Table 3), but varying the volume of a single aspiration cycle (0.5, 1.0, and 2 ml). In a first group of experiments, the same number of aspiration cycles (50) was adopted, as a consequence total volumes of 25, 50, and 100 ml, respectively were sampled. Fig. 2d reports the peak areas of each analyte in function of the aspired volume with a constant number of aspiration cycles. In spite of the increased volume sampled, β-pinene and isoamyl acetate peak areas decreased when the volume aspired in each cycle increased while for linalool peak area increased constantly.

The second group of experiments was run by adopting a constant total sampled volume (26 ml), and as a consequence varying the number of cycles (i.e. 52, 26 and 13) in function of the volume aspired in each cycle (0.5, 1.0, and 2 ml, respectively). Fig. 2e reports the peak areas of each analyte in function of the number of aspirations with a total volume sampled constant. From these results, it appears that for β-pinene and isoamyl acetate recovery increased when a higher number of cycles of lower volume was applied while the behaviour of linalool did not appear to be affected by number and volume of cycles. The aspiration volume adopted was also compatible with the β value

Table 4
Mean peak areas (%), repeatability and intermediate precision (R.S.D.) for the components characterizing rosemary headspace

No.	t_R (min)	Compound	SPDE			SPME		
			Repeatability		Intermediate precision	Repeatability		Intermediate precision
			Area (%)	R.S.D. (%)	R.S.D. (%)	Area (%)	R.S.D. (%)	R.S.D. (%)
1	4.29	α-Pinene	5.2	5.7	6.7	1.8	5.8	6.6
2	8.63	Limonene	4.5	8.7	9.8	1.5	9.1	10.3
3	9.20	1,8-Cineole	2.9	8.2	9.4	1.7	7.7	8.4
4	10.03	Isoamyl alcohol	2.9	7.2	8.6	0.1	10.2	10.8
5	16.62	Linalool oxide	1.1	6.8	7.2	0.7	6.4	7.9
6	18.05	Camphor	20.2	4.1	5.5	12.3	10.2	10.9
7	18.51	3,5-Octadien-2-one	1.6	8.5	8.9	0.8	11.6	11.8
8	19.56	Linalool	4.0	7.0	8.4	3.2	10.6	10.9
9	20.03	Bornyl acetate	25.4	6.6	7.1	33.2	11.3	11.6
10	22.64	Verbenone	15.6	9.3	9.6	28.0	12.4	12.9
11	22.95	Borneol	13.6	8.5	9.8	11.8	9.2	9.9
12	27.79	β-Ionone	0.4	9.6	9.7	1.5	7.1	8.1
13	32.68	Thymol	2.8	7.2	7.8	3.5	4.1	5.3
Total			100.0			100.0		

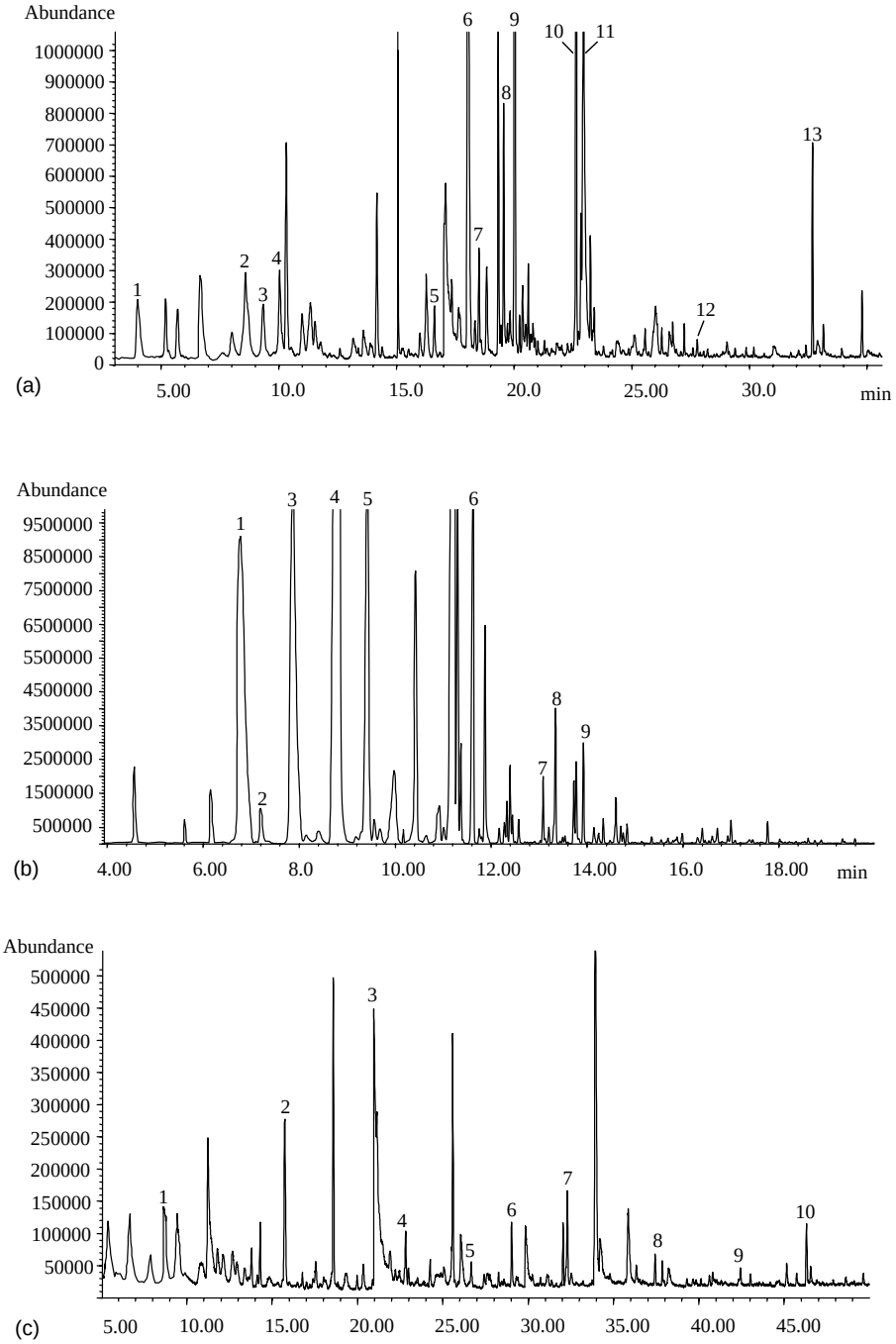

Fig. 3. (a) HS-SPDE–GC–MS profile of rosemary; oven temperature: from 20 °C (2 min) to 220 °C (5 min) at 5 °C/min. List of identified compounds: 1: β-pinene; 2: limonene; 3: 1,8-cineole; 4: isoamyl alcohol; 5: linalool oxide; 6: camphor; 7: 3,5-octadien-2-one; 8: linalool; 9: bornyl acetate; 10: verbenone; 11: borneol; 12: β-ionone; 13: thymol. (b) HS-SPDE–GC–MS profile of banana; oven temperature: from 0 °C (2 min) to 30 °C at 30 °C/min then to 200 °C (5 min) at 10 °C/min. List of identified compounds: 1: isobutylacetate; 2: ethyl butanoate; 3: 1,2-dimethylpropyl acetate; 4: isoamyl acetate; 5: isobuthyl butanoate; 6: isoamyl valerianate; 7: 1-methyl-hexyl butanoate; 8: hexyl butanoate; 9: isoamyl caproate. (c) HS-SPDE–GC–MS profile of green coffee; oven temperature: from 20 °C (2 min) to 40 °C at 3 °C/min then to 200 °C at 5 °C/min. List of identified compounds: 1: limonene; 2: 2-heptanol; 3: acetic acid; 4: benzaldehyde; 5: γ-butyrolactone; 6: furfuryl alcohol; 7: methyl salicylate; 8: phenylethyl alcohol; 9: isopropyl myristate; 10: *p*-vinylguaiacol. (d) HS-SPDE–GC–MS profile of red wine; oven temperature: from 20 °C to 220 °C (5 min) at 5 °C/min. List of identified compounds: 1: isoamyl acetate; 2: ethyl caproate; 3: isoamyl alcohol; 4: ethyl lactate; 5: 1-hexanol; 6: ethyl caprilate: 7: ethyl caprate; 8: ethyl succinate; 9: ethyl laurate; 10: phenylethyl alcohol; 11: ethyl palmitate. (e) HS-SPDE–GC–MS profile of white wine; oven temperature: from 20 °C (2 min) to 220 °C (5 min) at 5 °C/min. List of identified compounds: 1: isoamyl acetate; 2: ethyl caproate; 3: isoamyl alcohol; 4: 1-hexanol; 5: ethyl caprilate; 6: linalool; 7: ethyl caprate; 8: ethyl laurate; 9: phenylethyl alcohol; 10: octanoic acid; 11: decanoic acid.

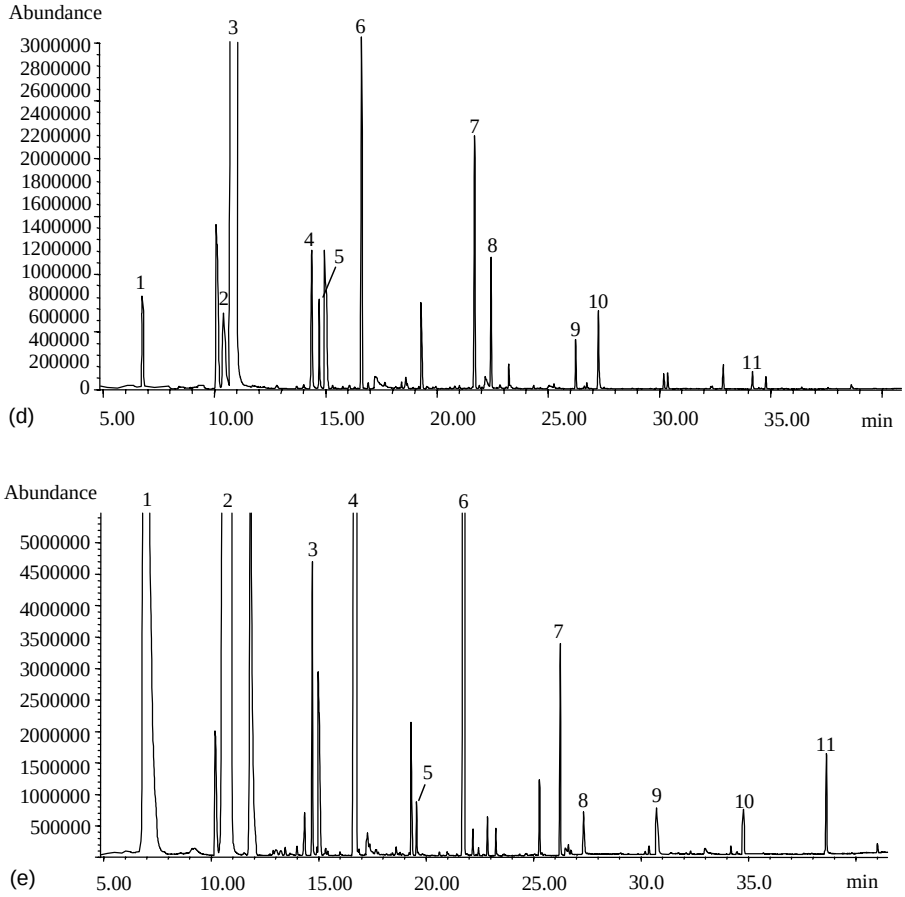

Fig. 3. (*Continued*).

arbitrarily chosen for these experiments since with a β of 9.6 together with aspired volumes higher than 1 ml, recoveries of some of the standard components decreased.

3.5. Helium desorption volume

HS-SPDE was applied to a series of standard samples to determine the most suitable helium desorption volume (0.5, 1.0, and 2 ml) under standardised conditions (Table 3). Fig. 2f reports the peak areas of each analyte in function of the helium desorption volume and shows that good recoveries for all analytes are achieved with 1 ml of helium; in particular this desorption volume gives the best recovery for β-pinene and isoamyl acetate.

3.6. Desorption plunger speed

This parameter was optimised by analysing a set of standard samples analysed under standardised conditions (Table 3) and desorption plunger speeds of 10, 15 and 25 μl/s. Fig. 2g reports the peak areas of each analyte in function of the desorption plunger speed and shows that a plunger speed of 15 μl/s affords a good recovery for all analytes, again, in particular for β-pinene and isoamyl acetate.

The last two parameters (helium desorption volume and desorption plunger speed) were carefully investigated because they significantly influence recovery and quality of the GC results. Musshoff et al. [11] found that the GC response increased with desorption volume, achieving a maximum at the full syringe volume (2.5 ml). Secondly, a plunger speed above 50 μl/sec was not compatible with the pressure control of the GC system. Moreover, too high a desorption plunger speed appeared not to permit a full desorption of each analyte from the PDMS into the helium stream completely. These authors therefore applied a relatively low plunger speed (10 μl/s) although this implied a relatively long desorption time, and, as a consequence, a low initial oven temperature (90 °C) to concentrate the desorbed analytes at the column inlet. We observed the same phenomenon, but probably because of the higher volatility of the fractions investigated a lower volume of helium (1 ml) and a higher desorption plunger speed (15 μl/sec) could be applied to desorb the analytes completely; the higher analyte volatility required a lower initial oven temperature (20 °C for all matrices with the exception of banana (0°)) to avoid chromatographic band broadening. In some cases, we observed a slight peak distortion of the most volatile components in spite of the low oven initial temperature.

From these experiments, it appears that with medium-to-high volatility and medium-to-low polarity analytes in aqueous media, good recoveries can be obtained (a) at moderately low temperature (50 °C) so that the partition equilibrium is in favour of adsorption into the polymeric coating, (b) with a moderate number of aspiration cycles (50) to avoid unwanted loss of the most volatile sampled components when the plunger returns to begin a new cycle, (c) with a relatively small aspiration volume (0.5–1 ml), (d) with a medium-to-low plunger speed (50 μl/s), (e) with an helium desorption volume of 1 ml, and (f) with a desorption plunger speed of 15 μl/s. These results are almost at all in agreement with those reported by Musshoff et al. [11] for amphetamines in hair samples of drug abusers; the differences were expected in view of the completely different nature of both analytes and matrices investigated.

HS-SPDE was then applied in combination with GC–MS to a number of matrices of interest for the nutritional field, in particular rosemary, banana, green and roasted coffee and red and white wines. The GC–MS results were compared with those obtained from the same matrices analysed by HS-SPME and S-HS samplings. Fig. 3 reports the HS-SPDE–GC–MS profiles of rosemary (a), banana (b), green coffee (c) and red (d) and white wines (e). Analysis conditions and a list of some of the characteristic components identified in each matrix are reported in the captions of each chromatogram.

HS-SPDE–GC–MS repeatability and intermediate precision were evaluated on rosemary. The same sample was analysed by HS-SPDE–GC–MS six times a day on three non-consecutive days. Table 4 reports the percent mean peak areas (%) and relative standard deviation (R.S.D.) for a group of 13 compounds identified in the rosemary headspace. HS-SPDE–GC–MS repeatability of the rosemary components was also compared to that of HS-SPME–GC–MS with a PDMS 100 fibre applied to the analysis of the same sample (Table 4). In both cases, the percent areas (%) of each component has been related to the sum of the 13 analytes peak areas under investigation to make the results easier to compare. Repeatability was good with both techniques; R.S.D.s of all components were comparable and ranged from 4.1% for camphor to 9.6% for β-ionone, in several cases HS-SPDE–GC–MS R.S.D.s are lower than those obtained by HS-SPME–GC–MS with the exception of 1-8-cineole, β-ionone, and thymol. HS-SPDE–GC–MS intermediate precision was also good; R.S.D.s were just a little higher than repeatability for all analytes investigated ranging from 5.5% for camphor to 9.7% for β-ionone.

The concentration capability of HS-SPDE was then determined and compared to that of HS-SPME by determining their concentration factors (CFs) calculated versus S-HS of a group of analytes characteristic of the matrices investigated. The CF of an analyte achieved by HS-SPDE or HS-SPME sampling techniques is the ratio between its areas obtained by HS-SPDE (or HS-SPME) and that obtained by S-HS, taken as 100. CF is not an absolute parameter because it closely depends on sampling conditions and on the physical state of the matrix, but it may be used for a relative comparison, provided that rigorous standard sampling conditions are applied. Here we report the HS-SPDE or HS-SPME CFs of some of the components characterising the roasted coffee headspace determined versus S-HS. Fig. 4 shows the GC–MS profiles of roasted coffee after S-HS, HS-SPME and HS-SPDE sampling techniques; the GC conditions are reported in the captions. Table 5 reports CFs achievable by HS-SPME and HS-SPDE calculated versus S-HS of a group of roasted coffee headspace components.

For all analytes investigated, CFs obtained by HS-SPDE are higher than those obtained by HS-SPME. In general, CFs increased with the volatility of the sampled analytes and were from three- to five-fold those of HS-SPME for the most volatile analytes, for instance for 2,6-dimethylpyrazine CFs by HS-SPDE is 1598 while with HS-SPME is 501, for 1-acetyloxy-2-propanone CFs by HS-SPDE is 1123 while with HS-SPME is 195. The only exception is *p*-ethylguaiacol, whose recovery is lower than that obtained by HS-SPME. This is probably due not only to its low volatility, which conditions the headspace/matrix partition, but also to its polarity and, as a consequence, to its solubility in PDMS. The higher CFs obtained with HS-SPDE than with HS-SPME can be explained by the fact that the volume of PDMS coating the needle wall is about eight times higher than that coating the SPME fused silica fibre (V_{SPDE}: 4.5 μl versus V_{SPME}: 0.6 μl). Several other polymeric coatings for SPDE needles, in addition to PDMS, are available in order to maximise recovery in function of the physico-chemical characteristics of the analytes investigated (PDMS/activated charcoal, PDMS/OV-225, PDMS/phenyl-methylpolysiloxane, PEG, DB-1701).

Table 5
CFs achieved by HS-SPME and HS-SPDE calculated vs. S-HS of a group of roasted coffee headspace components

No.	t_R	Compounds	HS	SPME	SPDE
1	8.67	Pyridine	100	161	1155
2	12.19	2-Methylpyrazine	100	189	1136
3	14.63	2,5-Dimethylpyrazine	100	577	1630
4	14.93	2,6-Dimethylpyrazine	100	501	1598
5	15.17	2-Ethylpyrazine	100	410	1453
6	17.00	3-Ethylpyridine	100	1425	3181
7	17.45	2-Ethyl-5-methylpyrazine	100	690	1708
8	21.36	1-Acetyloxy-2-propanone	100	195	1123
9	22.60	Furfuryl formate	100	149	784
10	24.26	Furfuryl acetate	100	371	1254
11	27.44	6,7-Dihydro-5H-cyclopentapyrazine	100	1908	1936
12	29.02	Furfuryl alcohol	100	224	1034
13	35.66	Guaiacol	100	787	1458
14	39.28	2-Acetylpyrrol	100	1211	1356
15	41.30	*p*-Ethylguaiacol	100	2919	2072

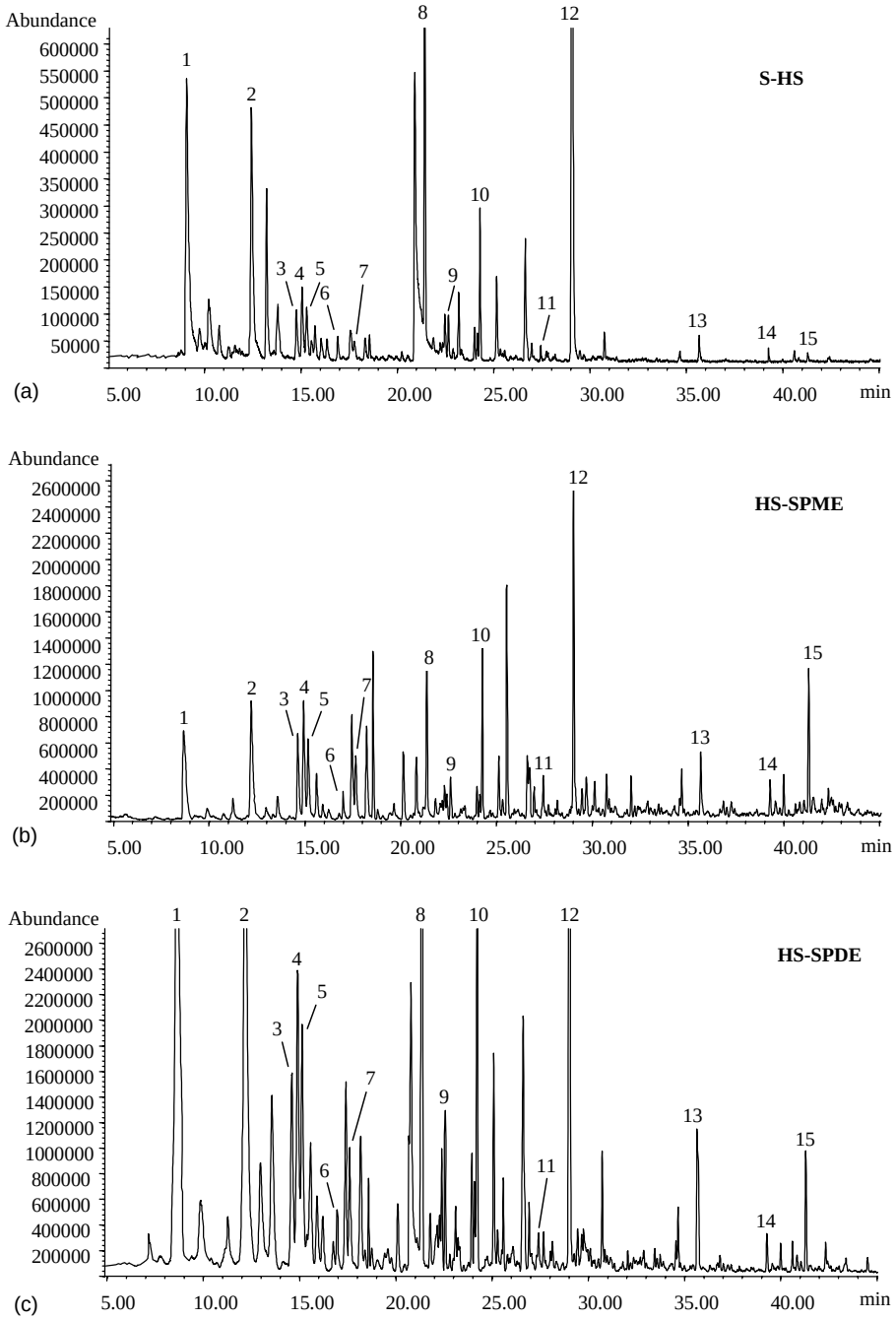

Fig. 4. GC–MS profiles of roasted coffee after S-HS (a), HS-SPME (b) and HS-SPDE (c) samplings. GC oven temperature: from 20 °C (2 min) to 40 °C at 3 °C/min then to 200 °C at 5 °C/min. (for further details see text; for component identification see Table 5).

4. Conclusions

Our results show HS-SPDE to be a further effective technique to bridge static and dynamic headspace techniques, because it gives high concentration factors like D-HS, and it is as easy to apply and to automate and as reproducible as S-HS. HS-SPDE has here successfully been applied for HS-sampling of a group of food matrices, giving good repeatability and intermediate precision for a series of components characterising the matrices investigated. Thanks to the volume of PDMS coating the syringe needle, HS-SPDE achieves a high concentration capability in particular for high-volatility analytes, and can therefore successfully be used for trace analysis.

On the other hand, further investigations are under way to understand from a theoretical point of view the influence of temperature and of number and volume of aspiration

cycles on the recovery of analytes with different polarity and volatility.

Acknowledgements

The authors are indebted with Chromtech (Idstein, Germany) for instrumentation and to T. van den Berg for technical support.

References

[1] Z. Zhang, J. Pawliszyn, Anal. Chem. 65 (1993) 1843.
[2] E. Baltussen, P. Sandra, F. David, C. Cramers, J. Microcol. Sep. 11 (1999) 737.
[3] B. Tienpont, F. David, C. Bicchi, P. Sandra, J. Microcol. Sep. 12 (2000) 577.
[4] C. Bicchi, C. Cordero, C. Iori, P. Rubiolo, P. Sandra, J. High Resolut. Chromatogr. 23 (2000) 539.
[5] C. Bicchi, S. Drigo, P. Rubiolo, J. Chromatogr. A 892 (2000) 469, and references cited therein.
[6] C. Bicchi, C. Iori, P. Rubiolo, P. Sandra, J. Agric. Food Chem. 50 (2002) 449, and references cited therein.
[7] G.E. Murphy, US Patent 5,565,622 (1996).
[8] M.E. McComb, R.D. Oleschuk, E. Giller, H.D. Gesser, Talanta 44 (1997) 2137.
[9] R. Eisert, J. Pawliszyn, Anal. Chem. 69 (1997) 3140.
[10] J. Lipinski, Fresenius J. Anal. Chem. 369 (2001) 57.
[11] F. Musshoff, D.W. Lachenmeier, L. Kroener, B. Madea, J. Chromatogr. A 958 (2002) 231.